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Fortification of Foods Established as Part of Nation's Nutrition Program

The principle of fortifying the human diet with vitamins and minerals which otherwise may not be supplied by typical diets in amounts required for adequate or optimum nutrition and good health has now become a practice fully recognized and accepted by government agencies and nutritional and medical authorities.

The history of mankind can be written in terms of man's eternal quest for food. Man's dietary habits throughout different areas and eras of civilization have both reflected and affected his way of life. In the course of the evolution of organized society, the solving of one nutrition problem frequently has automatically and simultaneously created new, and often more highly complex, nutrition problems.

Emergence of governmental policies which sponsor the supplementation of important staples of the diets of the most powerful nations with nutritive essentials which commonly are consumed in suboptimal amounts, constitutes a recognition and practical application of a basic truth of biology—that nutrition is the first line of defense in the battle for survival of the individual, of nations and of the species.

Nutrition research over a period of many years has carefully built up, piece by piece, a structure of facts concerning the chemical processes and substances involved in nutrition. As this structure has more nearly approached completeness, its broad outlines and significance have become more and more prominent to the public. Widespread popular interest and recent acceleration of efforts to apply findings of nutrition research to the regular dietary of the public of the United States already have resulted in an extensive program under which the public will enjoy a dietary supplemented or enriched with specific vitamins and minerals.

The Diet Problem Stated in Practical Terms

The chief diet problem in the quest of good nutrition up to the present has been the selection of the proper types of foods in proper amounts based on

knowledge of the nutritional values of foods. Lack of this knowledge by a large part of the population has made this an undependable procedure for assuring a well nourished population. It has been necessary therefore to resort to more scientific and readily applicable means, the supplementation or fortification of certain staple foods with vitamins and minerals to make up for deficiencies in commonly used foods. This constitutes the ultra-modern method of protecting the nutrition of all people.

Highly Organized Contemporary Life Creates New Variations of Basic Problems

The loss of inherent nutritional essentials from natural foods during processing and storage incident to the requirements for feeding concentrated masses in large cities distant from original food sources, is an important but remediable difficulty, characteristic of the differences between the way of life today and in simpler times. The food problems with which earlier civilizations were confronted did not include a scientific study nor an understanding of sterilizing and milling processes, food nutritional values, stabilization of keeping qualities, and the packing of foods in hermetically sealed containers.

Vitamin and mineral deficiencies of individual foods and attendant losses of nutritional essentials, such as the deficiencies of white flour and white bread which are the subjects of the first government sponsored enrichment program, are specialized cases of the central problem of the complete and balanced dietary, which are solved by specific application of the general principle of supplementation.

Food Fortification Is Achieved by Adding Nutritional Substances as Concentrates or in Pure Form

The use of concentrates prepared from natural food sources rich in vitamins or minerals for supplementing diets or for fortifying foods is now a well-established practice. Although the dietary and therapeutic use of chemically pure mineral compounds to prevent or correct mineral deficiency conditions has been practiced for a number of years, pure vitamin substances have been made available for such purposes only very recently by their isolation and synthesis. Many additional vitamins, available now for dietary purposes in concentrates from natural food sources, remain to be identified chemically before their synthesis will be possible and before they can be used as pure substances for fortifying foods.

Vitamin and Mineral Enrichment of Flour and Bread Urged as Health and Defense Measure

Inauguration of the flour and bread enrichment program was expedited as a public health, defense emergency measure, under the leadership of the Na-

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tional Research Council's Committee on Food and Nutrition. Although differing in detail, the program is similar in general purpose to the enrichment of all white flour bread in England decreed last year by the British Government as a war measure. The British enrichment program is mandatory and provides for the addition of vitamin B_1 and calcium. In the United States the enrichment program is urged but is not mandatory.

The immediate purpose of the program is to make available to the American public white flour and bread which contains amounts of vitamin B_1, nicotinic acid, and iron equal to or exceeding the amounts of these factors the bread would contain if it were made from "stone-ground flour from high vitamin quality wheat." It is required that all of these factors be present at prescribed levels if the bread is to be sold as "enriched bread". Addition of riboflavin is also mandatory after January 1, 1942. Vitamin D, calcium and phosphorus additions are encouraged.

The enriched bread may be made by adding correct amounts of the prescribed vitamins and minerals to the baking formula, by the use of high vitamin yeast, or by using special white flour which has been suitably enriched. The milling industry already is producing flour enriched to conform with the Defense Council committee's recommendations. Enriched bread made in accordance with the recognized procedures is on the market in many sections.

Specifications for the enriched flour and bread were announced to the public from Washington by the National Research Council's Committee on Food and Nutrition on January 29, 1941 (1). The committee pointed out that it is not to be assumed that eating only a slice or two a day of the new bread could have much effect on the adequacy of the diet. The committee emphasized the special importance of the "enriched flour" and "enriched bread" for those of the lower income classes who depend on bread for a large part of their energy food.

The term *enrichment* thus enters the nutrition vocabulary as a synonym for *fortification* to indicate the procedure whereby the amount of an essential food factor—vitamin, mineral, or other food substance—in a diet or in a specific food is increased by direct addition.

Addition of Milk Has Increased Nutritive Value of Baker's Bread

Enrichment of bread with milk proteins which enhance the nutritive value of wheat proteins, and with calcium and certain factors of the water-soluble vitamin B complex, by the addition of milk solids to the baking formula, for several years has been a standard and growing practice in the nation's baking industry. The use of milk in bread has been encouraged actively by the American Institute of Baking through its Department of Nutrition. As a consequence of the increasing use of milk in bread, average commercial white bread today contains

substantially higher levels of calcium than formerly, it was shown by analyses of bread from thirty-three bakeries in twenty-seven states conducted by that institute (2).

These studies indicated that six slices of average white bread per day will supply 30 per cent of the adult average minimum calcium requirement, and that a pint of milk in the diet each day along with the six slices of bread will supply approximately the entire adult calcium requirement.

The following material is quoted from the original report of the studies:

> . . . the average per cent calcium for the thirty-nine loaves, representing twenty-seven states, was 0.128 on dry basis and 0.080 on 38% moisture basis. The lowest figure found was 0.048% on a 38% moisture basis and 0.076% calcium on dry basis. The highest figure was 0.145% on a 38% moisture basis and 0.235% calcium on dry basis. The average ash was 1.86% on a 38% moisture basis and 3.00% on a dry basis. It is interesting to point out that the average per cent calcium found for white bread (38% moisture basis) is much higher than 0.048% calcium (dry basis) for wheat, as reported by Sullivan. . . . In the same article Sullivan reports 0.016% calcium (dry basis) for patent flour which gives some idea of the large increase in calcium of average white bread over the patent flour from which it is made.
>
> Due to the increased amount of milk being used today in the bake shop, and also to the use of yeast foods, the calcium content of commercial white bread on the market today is much higher than previously reported. . . .
>
> Ten control loaves with varying amounts of dry skim milk and dry whole milk were analyzed to determine the effect of milk solids on the calcium content of white bread. As the percentage of milk solids was increased, the percentage of ash and of calcium in the loaf also increased.

Vitamin D Irradiated Milk Pioneered in Showing Soundness of Foods Fortification Principle

Fortification of a basic food to enhance its value as a dietary source of a vitamin or nutritional substance—as fortification is understood in terms of modern nutrition—first was accomplished on a large and significant scale by the commercial irradiation of milk with ultraviolet light to provide a dietary source of vitamin D. The first commercial production and distribution of irradiated vitamin D partially defatted milk, in a dry form, in this country began in January, 1927 (3) under the direction of Dr. G. C. Supplee, who is in charge of The Borden Company Biological and Chemical Research Laboratories at Bainbridge, N. Y. The first vitamin D irradiated milk for the fluid milk market was offered to the public in this country in 1933, by The Borden Company fluid milk division in Detroit, Michigan. The broadest, most extensive use of irradiation for elaborating vitamin D in foods has been made by the evaporated milk industry, which since 1936 has irradiated virtually the entire output of its product. Since the daily users of evaporated milk throughout the world number in the hundreds of millions, a significant measure of the world-wide, popular appreciation of vitamins in nutrition may be attributed to the vitamin D enhancement program of the evaporated milk processors.

Vitamin D irradiated milk was a practical development of the discovery in 1924 independently by Hess (4) and Steenbock (5) that ultraviolet rays will produce vitamin D properties in certain food substances. Commercial application of the irradiation process to milk resulted from a series of research investigations in which Supplee, Hess, and a number of other workers were associated. Coordination of the findings of various workers by the early nineteen-thirties, facilitated the orderly commercial production, by license under the Steenbock patent, of vitamin D irradiated milk throughout the country. Vitamin D milk also is produced by methods other than irradiation which include the feeding of vitamin D activated yeast to cows, or the direct addition to milk of natural, irradiated, or activated vitamin D materials.

The principle of imparting enhanced vitamin D activity to fluid milk, evaporated milk, dried milk, and special milk products for infant feeding is now widely applied commericially. The value of irradiated milk products as outstanding food sources of vitamin D is universally recognized by the medical profession and by nutritionists. It is thus apparent that the commercial production of vitamin D milk pioneered the present manufacture of foods supplemented with various vitamins and minerals.

Irradiation Work Foreshadowed Mass Application of Laboratory and Clinic Supplemented Diets

The importance of the application to the diets of entire portions of the population of nutritional practices established as valuable in laboratory and clinic, through the mass supplementation of a basic food, was stated 14 years ago in a report by Supplee and Dow (3) on the potentialities discerned then in irradiated milk. The following statement, quoted from the original paper, essentially states the general principle which is now achieving wide recognition:

> The antirachitic activation of milk to a degree that renders it a reliable prophylactic material for diets used in the prevention of rickets is a subject that presents manifold possibilities. . . . Up to the present time the feeding of irradiated milk has been confined to hospitals and institutions in which equipment and personnel are available for the proper preparation of the product. Such limitations will of necessity prevent a wide dissemination of the benefits known to result from the use of activated milk and, unless the principle is applied to a suitable and easily accessible preparation, many infants will be deprived of a needed prophylaxis of rickets. While irradiated dry milk has been prepared experimentally for clinical observations, it has only been recently that the principle of irradiation has been utilized for mass production of this product.

Vitamin B_1 Fortified Dried Partially Defatted Milk Among First Supplemented Foods

Current recognition of the need of enriching foods by the addition of vitamins was initiated by problems arising from the artificial feeding of infants. The

The amino acid compositions of the two chief proteins of milk, casein and lactalbumin, are shown in Table 1. To their amino acid makeups—particularly their complements of the 10 indispensable amino acids—may be attributed a large measure of the remarkable nutritive qualities of these two milk proteins—for maintenance of the adult as well as for the growth of the young for which they are primarily synthesized by nature.

The 22 known amino acids, the milk protein contents of which are shown in Table 1, are the building units from which, with possible individual omissions, are constructed all natural food proteins, whether of animal or vegetable origin. Of these 22 amino acids, the 10 which are classified as "nutritionally indispensable" are specially identified in the table. Various aspects of the subject of the 10 indispensable amino acids will be discussed more fully in subsequent paragraphs. Of primary significance at this point is the fact that the presence, absence, or quantitative proportions of these 10 amino acids in a dietary protein largely determine its biological value.

Amino Acid Compositions of Milk Proteins Make Casein and Lactalbumin Biologically Complete

Either of milk's proteins, casein and lactalbumin, alone, or both together, because they provide all 10 of the indispensable amino acids, will give satisfactory growth and reproduction in test animals, when fed in sufficient amounts along with an otherwise adequate diet. For this reason, the milk proteins are classified as "biologically complete" proteins.

However, the observations of Mendel (2) in analyzing the results from the classical investigations of Osborne and Mendel into the nutritive values of proteins initiated in 1911, typically exemplify one of the basic reasons for the nutritive superiority of "milk proteins":

> . . . The presence of lactalbumin along with casein in milk furnishes a mixture of proteins which is preferable, gram for gram, to casein alone. The relative amino-acid shortcomings of the casein, as exhibited in the low content of cystin, are averted by the lactalbumin. From the point of view of economy, therefore, it is advantageous to learn the amino-acid make-up of all available food proteins, so that they can be exhibited in proportions furnishing a balanced total amino-acid make-up as nearly ideal as possible. . . .

Individual proteins are made up of various combinations of some or all of the 22 amino acids. Many proteins, however, do not contain all of these amino acids, and some are deficient, especially in one or more of the 10 essential amino acids. Proteins which are lacking in one or more of these indispensable 10 are classified as "biologically incomplete" proteins because as sole protein components of an otherwise adequate diet they will not permit normal growth and body health. Incomplete proteins are found among those from both animal and plant sources.

TABLE 1.

PERCENTAGES OF AMINO ACIDS IN MILK PROTEINS

(The first 10 amino acids, printed in all capital letters, are "nutritively indispensable")

AMINO ACIDS	CASEIN	LACTALBUMIN	LACTALBUMIN AND CASEIN (As Found in Milk) *
ARGININE	3.79	3.0	3.6
HISTIDINE	1.81	2.1	1.9
ISOLEUCINE }	9.7	14.0	10.3
LEUCINE }			
LYSINE	6.20	8.8	6.6
METHIONINE	3.10	2.8	3.1
PHENYLALANINE	3.88	1.3	3.5
THREONINE	3.50	4.70	3.67
TRYPTOPHANE	1.11	2.7	1.4
VALINE	7.9	3.3	7.3
Alanine	1.9	2.4	1.9
Aspartic Acid	4.1	9.3	4.8
Cystine	0.37	3.1	0.7
Glutamic Acid	21.8	12.9	20.5
Glycine	0.5	0.4	0.5
Hydroxyglutamic Acid	10.5	10.0	10.5
Hydroxyproline	0.2	.....	.....
Proline	9.0	3.8	8.3
Serine	0.5	1.8	0.7
Tyrosine	6.01	2.0	5.4

NOTE.—All values in the first two columns of this table are those given by Calvery in the Schmidt treatise (5) except those for threonine of casein and lactalbumin (6), for arginine, histidine, lysine, methionine, tryptophane, and tyrosine of casein (7), for phenylalanine of casein (8), for methionine of lactalbumin, and for cystine of casein and of lactalbumin (9). Research on amino acid compositions of milk proteins and other food proteins is not yet complete; values for norleucine (usually included in leucine and isoleucine values), for hydroxyvaline, for citrulline, and for thyroxine—one or more of which have been included by various workers in lists of amino acids—are not given in the above table.

*Values in third column are estimated total percentages of the amino acids in casein and lactalbumin together, at approximately 2.95% and 0.5%, respectively, as found in milk.

The nutritively complete character of the proteins of milk, however, has been established by biological feeding tests and by chemical analyses which show that casein and lactalbumin contain all of the essential amino acids and, in addition, nutritively valuable amounts of virtually all the other amino acids.

Significance of Term Indispensable *Applied to Amino Acids in Diets*

The indispensability of the 10 amino acids that are classified as nutritively essential has been demonstrated by feeding experiments in which combinations of pure amino acids, rather than natural proteins, were fed to the animals as the sole source of protein material in the diet.

In many long and carefully controlled tests of this sort, nutrition research workers found that when any one of 12 of the 22 amino acids was withdrawn from a satisfactory diet, growth, health, reproduction, and other physiological processes or functions proceeded satisfactorily. These 12 amino acids, therefore, were classified as "nutritively dispensable".

In contrast, when one or more of the other 10 amino acids were not provided in the diet, several or all of these normal life processes were interrupted and the animals could not grow, reproduce, or maintain health, and they frequently died. It became apparent, therefore, that these 10 amino acids were "nutritively indispensable"—that they must be supplied in the diet for normal health.

However, in the light of the rapid advance and new techniques of the medical and biochemical sciences it is unsafe to take for granted that optimum nutrition or even continuance of maintenance of proper nutrition would prevail in the absence of all or some of the now designated "nutritively dispensable" amino acids contained in natural food proteins. Future investigation may reveal specific and enhancing functions of one or more of the "dispensable" amino acids for the prevention of insidious deficiency diseases—which would necessitate a more up-to-date reclassification of such a factor as "indispensable".

How the animal body can get along without certain of the amino acids became understandable when it was revealed by chemical analysis that proteins from tissues of animals fed only the 10 essential amino acids contained various amounts of other amino acids, although these had not been fed. It was evident that animal bodies, within certain limits at least, are able to build up these 12 dispensable amino acids by their own processes of metabolism if the 10 essential amino acids are supplied in the diet. Such body synthesis of appreciable amounts of the indispensable 10 amino acids, however, proved to be impossible.

Amino Acid Feeding Experiments Described by Nutrition Workers

Essential facts now known concerning the dispensability and indispensability of the various amino acids as dietary components, are stated by Rose and Rice (3):

> In previous publications from this laboratory . . . it has been shown that only ten of the twenty-two amino acids known to exist in proteins are *indispensable*

dietary components. These are tryptophane, lysine, histidine, phenylalanine, leucine, isoleucine, threonine, methionine, valine and arginine. With the exception of arginine, the removal of any one of these compounds from the food leads to a profound nutritive failure, accompanied by a rapid decline in weight, loss of appetite and eventual death. On the other hand, the exclusion of arginine from the ration is followed by much less pronounced effects. The subjects continue to gain, but at a suboptimal rate. This is accounted for by the fact that arginine, in contrast to the other members of the indispensable group, can be manufactured by the cells, . . . but at a speed which does not quite keep pace with the demands of *normal* growth. The twelve remaining amino acids are dispensable in the sense that they can be synthesized in adequate amounts out of materials ordinarily available in the organism.

In arriving at the above conclusions young rats served as the experimental animals. Consequently, it does not follow necessarily that the findings are applicable to other species. . . . It becomes of importance, therefore, to ascertain . . . whether other mammals also manifest the ability to thrive on relatively simple mixtures.

The nutritive role of the individual amino acids has now been established for the adult dog. This was undertaken as a preliminary to extensive investigations on the maintenance of nitrogen equilibrium in various species by oral and intravenous alimentation. . . .

Three dogs were employed in determining the physiological importance of the other amino acids of the indispensable group. Invariably, the removal from the food of any one of these compounds was followed by a pronounced negative nitrogen balance. Furthermore, the restoration of the missing amino acid to the diet uniformly resulted in a positive nitrogen balance. These data demonstrate that *the qualitative amino acid needs of the dog are identical with those of the rat.* The fact that two widely different species require for their well-being the same components of the protein molecule, increases the probability that other mammals, including man, may manifest like responses.

Milk Proteins Supplement Amino Acid Deficiencies of Other Proteins, and Are Accompanied by Valuable Minerals and Vitamins

Milk proteins are of unusual nutritive value because the particular amino acid deficiencies of various other proteins commonly included in typical diets are supplemented efficiently by the relatively high amounts of certain amino acids in casein and in lactalbumin, and also because associated with milk proteins are certain biologically important mineral and vitamin substances which frequently do not accompany other proteins.

Of especial importance in the economy of practical diet planning is the ability of milk proteins, even in relatively small amounts, to increase the nutritive efficiency of grain proteins, or of other less complete proteins, which has been established by the experiments of a number of workers. Some of these experiments now are classics in the literature of nutrition research, and others are still in progress and promise to explain more fully the biological chemistry of all proteins—which have been called "the essential stuff of living tissue".

Most proteins are composed chiefly of carbon, hydrogen, oxygen, nitrogen, and sulphur. Phosphorus and small amounts of other elements, including iron, are also present in some proteins. Comparative analyses of typical proteins show variations of less than 4 per cent in the ultimate elementary composition of proteins (4). The variations among common proteins in amino acid composition, however, extend over a wide range.

The wide range of differences in nutritive value of various proteins in the diet, the primary importance of adequate proteins in nutrition, and the relatively higher cost of good protein foods, have combined to stimulate efforts to evaluate the nutritive efficiency of proteins economically available for diets of men and of livestock.

Tables of nutritive values assigned to proteins by different workers employing the several assay methods and criteria recently have been compiled by McCollum and his associates (1). The tables include values for proteins of cereals, legumes, and vegetables, and for proteins of milk, eggs, and meats. These authors summarize the collective significance of the extensive literature relating to the nutritive value of various classes of proteins:

> In general . . . proteins of animal origin have a higher nutritive value than those of any other class. This is understandable, particularly in the case of milk and eggs, since these proteins must provide all the amino acids required by the young mammal or developing chick. . . .
>
> As is indicated by the tables, all cereal proteins have lower nutritional values than those, in general, of meat and animal products. . . .

Amino Acid Compositions of Milk Proteins Compared with Amino Acid Compositions of Wheat Proteins

The nutritive superiority of such proteins as casein and lactalbumin of milk, over various proteins of plant origin, including some of the cereal grain proteins, is due to the presence in relative abundance in milk proteins of the essential amino acids, and the absence or comparative deficiency of one or more of these amino acids in the inferior plant proteins.

These differences in amino acid contents are illustrated in Table 2.

Use of Milk in Experimental Diets Demonstrates Supplementary Values of Proteins

Of more significance than the nutritive value of individual proteins is the ability of certain proteins to supplement and thereby enhance the nutritive value of other less nutritious proteins, as McCollum (1), who was among the pioneer workers to investigate supplementary relationships of proteins, has emphasized.

TABLE 2.

COMPARISON OF CONTENTS OF 10 ESSENTIAL AMINO ACIDS OF MILK PROTEINS AND WHEAT PROTEINS

ESSENTIAL AMINO ACIDS	MILK PROTEINS		WHEAT PROTEINS		
	CASEIN	LACTAL-BUMIN	GLIADIN	GLUTENIN	GLUTEN
Valine	7.9	3.3	3.4	0.2	.....
Leucine and Isoleucine	9.7	14.10	6.6	6.0	4.1
Phenylalanine	3.88	1.3	2.4	2.0	1.0
Tryptophane	1.11	2.7	1.1	1.7	.....
Threonine	3.50	4.70	*	*	*
Methionine	3.10	2.8	2.1	.....	.....
Arginine	3.79	3.0	2.9	4.7	4.4
Histidine	1.81	2.1	1.5	1.8	1.2
Lysine	6.20	8.8	0.6	1.9	2.2

*Value not determined.

NOTE.—All values for wheat proteins in this table are those given by Calvery (5). Sources of values for milk proteins are as noted with Table 1.

An early example of the remarkable values of milk proteins in supplementing deficiencies of other proteins is described by Steenbock and Hart (10) also among the pioneer workers to study this subject:

> . . . How often we see or hear the statement "It is high in protein, the great blood and muscle building food." To be sure without protein these tissues cannot be built, but we now know as a result of work in our experiment stations that proteins differ tremendously in the extent to which the animal can use them for these purposes. For instance, if two animals as near alike as possible in ancestry, age, size, and vigor, are given one hundred (100) pounds, respectively, of the protein of milk and the protein of the ordinary grains, there will be retained sixty-five (65) pounds of milk protein, but less than thirty (30) pounds of the grain protein in spite of the fact that they are both well digested and absorbed. These proteins are so different in composition that they cannot be utilized equally well for building body tissues. This in itself would not be a serious matter if the animal could advantageously eat so much more of the cheaper grain protein, but its capacity to do this is so limited that growth and development suffer. Mixtures of grain proteins with milk proteins are as efficiently retained as milk protein itself since milk contains in highly concentrated form the essential substances. What grain protein lacks is supplied abundantly by the milk.
>
> Even the small amount of protein in whey supplements grains excellently. In the Wisconsin experiments with growing pigs, where this by-product of the cheese

industry was fed at the rate of four (4) pounds daily as a supplement to a corn meal gluten feed ration, 47 per cent of the total proteins fed were stored by the animals. Without the whey proteins, which were but 16 per cent of the entire protein mixture, only 30 per cent of storage resulted. These figures are significant. Certainly our stockmen cannot afford to allow whey to be run into the gutter or creek to turn into a vile smelling waste product. It should be saved and fed to the stock.

Milk in any form, whether whole, skimmed, or as buttermilk or even whey, and as the important product cheese in all its different forms, is a superior source of protein. This is not because of the amount of protein—for the milk products listed differ too widely—but because the protein seems to be just what the body needs to build and repair its tissues.

What applies to milk protein in a large measure also applies to meat and egg proteins for they have helped enormously in the history of mankind in maintaining the human race under otherwise unfavorable conditions. But at present and, no doubt, as she will in the future, the dairy cow stands in a class by herself as an economical producer of excellent protein. This is true because she, of all animals, can use to best advantage the proteins of roughage such as hays, corn stover, and silage which are too bulky for human food.

Milk Was Basic Food in Pioneer Protein Studies

Among the classic pioneer experimental work upon which was founded much of the present understanding of the significance of amino acids composition in the nutritive values of different proteins—and the supplementary values of milk proteins—were studies conducted over a period of years by Osborne and Mendel, the general significance of which is summarized in the words of Mendel (2):

> T. B. Osborne and I . . . were able to facilitate the study of the role of the individual proteins and amino-acids in growth by introducing in the artificial ration the use of what we have termed "protein-free milk."
>
> This consists of the dried residue of milk after removal of fats and proteins. . . . On mixtures of "protein-free milk," sugar, starch, and purified fats along with selected isolated proteins, young white rats (and in some cases mice . . .) have grown to maturity and have in turn produced young even in the third generation. . . .
>
> In the case of some . . . proteins, notably gelatin, zein, gliadin and hordein, an explanation for the failure of growth was at once suggested by the known deficiencies of each of these substances in respect to one or more amino-acids already ascertained to be yielded by the adequate proteins. The tryptophan group (with its indol nucleus) is missing in gelatin and zein; the diamino-acid group of lysin is lacking, or nearly so, in zein, gliadin, and hordein. Other shortcomings are the lack of the tyrosin group in gelatin and the glycocoll group in zein. . . .
>
> If we analyze the situation as revealed in the charts of some actual experiments it becomes apparent that both lysin and tryptophan are unquestionably necessary as constructive units in growth. The decline brought about by the zein food can be stopped by the addition of tryptophan, as such, to the diet. This results in maintenance; but no growth ensues until lysin also is added . . . .

The significance of other amino-acids derived from proteins needs to be studied in comparable ways. Experimentally such studies are complicated by the fact that most available proteins are not entirely devoid of the important amino-acid nuclei. The supply of the missing amino-acids need not be in the form of the isolated compound. Suitable proteins which yield them answer in the same way . . . .

The possibility of growth and the extent to which it is accomplished are limited by the supply of each essential amino-acid. It matters not whether this is exhibited as such or in the guise of protein; in either event the "law of the minimum" is exemplified. The amino-acid shortcomings of one protein can be made good by supplementing it with another protein in which they do not exist to the same degree.

Let us consider what these new observations mean for the problem of protein minimum. It may now be preferable to speak of amino-acid minima. The differences between proteins appear in a new light. . . .

Superiority and Biological Completeness of Milk Proteins

Milk proteins are unique in that they are biologically complete, providing all the amino acids required for growth, maintenance and reproduction.

The fundamental early work, such as that by Osborne and Mendel, and others, establishing the fact that certain amino acids must be supplied by the dietary protein, has been verified and extended by recent feeding experiments wherein mixtures of pure amino acids were used in the diet in place of proteins; data from such recent investigations are described by Rose and one of his associates (3) quoted elsewhere in this discussion.

Pertinent examples of the pioneer investigations of Osborne and Mendel, illustrating the nutritive superiority of milk proteins and their complementary value in supplementing proteins from other sources follow:

> Satisfactory growth was maintained on a diet in which the milk protein casein was the whole protein; only a little more than maintenance of body weight resulted from gliadin (one of the proteins of wheat) as the sole protein, and loss of weight resulted with zein (a protein of corn) as the single protein constituent of the dietary.
>
> The nutritive decline resulting from zein was reversed and considerable growth prevailed when the zein was supplemented with tryptophane and lysine.
>
> Zein as the sole protein of the dietary, while incapable of maintaining animals, supported good growth when supplemented with 4.5% of the more "perfect" milk protein, lactalbumin—although this amount of lactalbumin alone was insufficient to sustain a normal rate of growth. These results illustrate the ability of the "complete" protein (lactalbumin) to complement the amino acid deficiencies of an "incomplete" protein (zein).

Satisfactory growth was obtained with 18 per cent of the milk protein casein as the only protein in the diet, and with 18 per cent of edestin (hempseed protein), contrasted with reduced growth when these protein levels were cut to 9 per cent. In the first instance the addition of cystine to the 9 per cent casein permitted resumption of good growth and the addition of lysine to the smaller amount of edestin similarly accelerated growth.

These results demonstrated that a relative deficiency of one or more of the amino acids may limit growth, even in the presence of an abundance of protein *per se*—and that the supplementing of a dietary protein supply with an increased amount of one or more of certain amino acids may accelerate growth.

These brief illustrations also serve to demonstrate that even the breaking up of a natural protein system such as that of milk, into its fractions separable by virtue of differences in chemical properties, does not significantly degrade the nutritive character of the specific protein component. On the other hand, however, optimal nutritive and biological responses are to be expected when a protein food such as milk is utilized *in toto* and without disturbance of the normal balance of physico-chemical relationships prevailing in the natural product. In practical nutrition this beneficial result is accomplished by natural foods containing their quotas of specific proteins—the supplementing and complementing of particular amino-acid-deficient proteins being most effectively and economically achieved by milk and dairy products.

Nutritionally "Dispensable" Amino Acids Also Are Biologically Valuable

Vast ground remains to be covered before knowledge of the chemistry of the proteins and the physiological functions of their amino acids may be regarded as approaching completeness.

That the division of the amino acids into two groups, nutritionally "dispensable" and nutritionally "indispensable", although extremely useful and based on sound principles of biological chemistry, may be confusing unless fully understood, has been pointed out by various authors (4, 11). The fact that a number of amino acids can be synthesized within the body if the diet supplies adequate amounts of other amino acids which cannot be so synthesized, does not in any way imply that "dispensable" amino acids are to be regarded as nutritionally superfluous. Unless present in the diet in disproportionately large quantities, such amino acids usually are physiologically valuable—to the extent that, as in the case of cystine, they may be mistakenly regarded as indispensable. Cystine for many years was looked upon as the sole and essential sulphur-containing protein constituent; this misconception was strengthened by the demonstrable supplementary growth promoting value of cystine when added to restricted protein diets.

The isolation of methionine from casein and from other proteins (12), and experimental evidence produced by Rose *et al* that it, rather than cystine, is the nutritionally essential sulphur-containing amino acid is an example of how a generally accepted view regarding the very complex physiological chemistry of the proteins may be invalidated by new and frequently unexpected facts obtained by research. An earlier tendency to look upon comparatively limited amounts of cystine in some proteins as an example of perhaps undue frugality of nature, ultimately may have to be revised if it is established that limits on the amounts of cystine occurring in natural proteins may reflect possible danger of disruption of certain functions of intermediary metabolism when fed in excess or out of balance with other food substances.

Protective Action of Milk Casein Against Organic and Tissue Injury Exemplifies Amino Acid Biochemistry

Only during the last few years has physiological chemistry been able to determine and demonstrate experimentally the effects of individual amino acids on tissues and organs. This recent achievement facilitates a preventive pathology where amino acid malnutrition can be shown to affect tissues or organs adversely, and makes possible in practical nutrition a step toward the protection of tissues by the application of nutritional knowledge of the amino acids, and toward the choice of dietary proteins for their tissue-protecting qualities.

Damage to liver and kidney tissues resulting from a disbalance of the metabolism and utilization of the sulphur-containing amino acids, possibly interrelated with vitamin deficiency, as well as with fat and carbohydrate metabolism, is one of the current subjects of research pertaining to the broad subject of amino acid functional interrelationships.

Since cystine and methionine have served as examples elsewhere in this discussion, and since casein has been shown to exhibit protective qualities in experimental diets, some of the essential observations or deductions from research pertaining to these several interrelated subjects will be presented here briefly as items typifying the growth of knowledge of the functions of the amino acids; some additional information regarding the metabolism and the biological value of the amino acids of casein also will be included.

Significance of these data, regarding the protective qualities of milk proteins, casein in particular, from the research of many workers, which will be referred to specifically in subsequent paragraphs, may be summarized briefly and in general as follows:

1. The proteins of milk as a whole, or the single milk protein casein, in the diet tend to protect against cirrhosis and fatty degeneration and infiltration of the liver; against hemorrhagic degeneration of the renal (kidney) cortex, and against cystinuria (cystine in the urine)—all of which pathological conditions apparently may result either from a rela-

tive excess of cystine, or from a relative deficiency of choline in the diet. This protective effect is attributed largely to the presence in milk proteins of methionine from which the body may synthesize the choline or choline component radical required for the metabolism of cystine.

2. Casein in the diet reduces the incidence of artificially induced liver cancer in experimental animals.

3. Hyperplasia in the forestomach and peptic ulcer experimentally and dietically induced in test animals also have been prevented by the proteins of milk, both casein and lactalbumin—particularly the former—introduced in the experimental dietary.

Recent Research Findings Concerning Amino Acids Contained in Milk Protein, Casein

Brief references to individual data summarized in the preceding paragraphs follow:

It has been shown that cystine is not an essential amino acid in the diet—that it can be synthesized within the body from dietary methionine (13).

Methionine is as efficient as cystine for promoting growth (14, 15, 16, 17, 18).

Methionine is an essential amino acid, independent of its relationship to cystine (19).

Methionine cannot be synthesized within the body from cystine (20).

In comparison of the effect of the two amino acids on growth and lactation, it was reported that added cystine did not increase growth promoting properties of peanut meal protein, but did cause a slight increase in lactation promoting properties; methionine, however, improved growth and lactation (21).

Methionine and cystine, when fed as constituents of casein and lactalbumin, were catabolized both qualitatively and quantitatively in the same way as when they were administered in the form of free amino acids in cystinuria (9).

Addition to experimental diets of choline, a basic constituent of lecithin, recently classified as a factor of the vitamin B complex (22, 23), was found effective in the prevention of fatty livers (24) and it was shown that one gram of casein is equivalent in its preventive action on liver fat deposition to 7 to 8 mg. of choline (25).

The lipotropic action—the property of preventing abnormal accumulation of fat in the liver—of various proteins in the order of decreasing

intensity were listed as follows: Gromax and whale muscle protein, casein, albumin, beef muscle protein and edestin, fibrin, gliadin, gelatin, and zein (26).

The administration of methionine with cholesterol-containing diets markedly decreased the deposition of cholesterol in the liver in both the free and ester form; small amounts of cystine increased the deposition of glyceride in cholesterol fatty livers (27).

Experiments with different proteins in the basal diet suggested that proteins high in methionine and low in cystine were protective against renal degeneration (28).

Dietary casein exerts lipotropic action on fat deposition caused by cystine; the preventive action of methionine on liver fat deposition on an 8% casein diet increased until the amount of added methionine was 0.2% of the ration (29).

The lipotropic effect in rats on a low protein (5% casein), high fat (40% lard) diet supplemented with methionine and cystine to the extent that the sulphur amino acid content was equivalent to that of a 20% casein diet was found to be similar to that observed when the level of the casein in the unsupplemented diet was increased to 20%. Fatty livers were obtained when the casein content of the unsupplemented high fat diet was decreased to 15 or 5% (30).

The total lipid content of rat livers was found to be 77.5% less when a 5% gliadin diet containing 40% lard was supplemented with methionine, than when no methionine was present in the diet (31).

In a study of the phospholipid metabolism, methionine, cystine, and cysteine were found to accelerate phospholipid turnover in the liver (32) whereas glycine, alanine, serine, tyrosine, proline, glutamic acid, and asparagine were inactive (33).

Choline was found to be a dietary essential if a ration contained less than 0.8% methionine; the choline requirement varied inversely with the dietary methionine and was increased by dietary cystine (34).

Casein was the most protective of the proteins used in preventing hemorrhagic degeneration in young rats; the degeneration was absent or very slight in a low choline diet containing more than 0.8% of methionine, regardless of whether this level was attained by supplementary methionine or by protein levels high in casein, lactalbumin, or fibrin (34).

Methionine was found to furnish the methyl group of choline and creatine (35).

In the absence of added choline, 26% of casein was necessary to prevent renal lesions, and 34% to prevent fatty livers (36).

Cirrhosis of the liver which developed on high fat (55 and 70% fat) low protein (10% casein) diets was prevented by the addition to the diets of choline hydrochloride at levels of 40 to 60 mg. per rat per day, alone or in combination with methionine or cystine; addition of methionine at level of 25 mg. per rat per day indicated slowing of cirrhotic process; addition of cystine at 25 mg. per rat per day did not retard development of cirrhosis (37).

Simultaneous addition of casein and riboflavin to the diet substantially protected rats from liver cancer induced by feeding methyl yellow; addition of either casein or riboflavin alone reduced the incidence of liver cancer from 100% to 70 to 80%; addition of both together reduced incidence to 3% (38).

The etiology of gastric ulcer has been the object of manifold investigations for many years. Hyperplasia in the forestomach of rats and peptic ulcer induced by experimental dietaries have been generally reported to result from low protein diets (39, 40, 41, 42, 43, 44, 45).

Conversely, increased protein in the diet, especially the more biologically complete proteins, has been shown to reduce or prevent the incidence of the gastric lesions. Some investigations (45) attribute this protective action of particular proteins to the biologically and nutritively superior quality whereas others (44) ascribe the beneficial effects to the interrelation of the protein and vitamin supplementation.

Matzner, *et al* (46) report their conclusions concerning the effectiveness of proteins in preventing gastric lesions as follows:

> Proteins in the form of casein, gelatin, elastin and lean meat afforded a marked degree of prevention in contrast to the carbohydrate and fat groups.
>
> The mechanism of this protection is ascribed to the interference of proteins with proteolytic activity of pepsin, which may be due to a competitive retardation of digestive products or the acid combining power of proteins or the combined effect of both actions.

Unpublished data obtained at the Borden Biological and Chemical Research Laboratories (47) have shown that gastric ulcers and hyperplasia of the forestomach of rats are induced experimentally with high regularity by a diet consisting of substantially 90% white flour, minerals, fat, and natural or synthetic supplements of the vitamin B complex. However, when as little as 10% edible casein or lactalbumin separated from milk is introduced in the dietary the incidence of the gastric lesions is markedly reduced or entirely prevented. The vitamin supplementation with factors of the vitamin B complex appears to exert only a secondary effect on the incidence of hyperplasia and gastric ulcers, but does have a markedly favorable effect upon the growth and physical condition other than the localized stomach lesions.

The results from the numerous experimental studies interrelating the beneficial effects of proteins, and particularly milk proteins, for the prevention

of stomach lesions in experimental animals, are in general conformity with widely recognized medical practice for the treatment of peptic ulcers in the human subject, where milk plays a foremost part in the therapeutic dietary regimen.

Unusual Mineral Combinations—with Phosphorus and Calcium—Enhance Over-All Nutritive Value of Casein

Beyond the favorable amino acid make-up of the proteins of milk, is a notable aspect of the chemical composition of casein, namely its content of approximately 0.8% organic phosphorus exclusive of nucleic acid or lecithin. Casein thus is one of the few familiar proteins which may be classified as a *phosphoprotein*. Approximately 25% of the phosphorus of whole milk is present in the casein. Referring to some of the literature concerning the phosphorus of casein, McCollum and associates (1), cite authors (48, 49) who believe that the particular chemical properties of casein are such that maintenance of a low phosphate concentration in the intestinal tract facilitates absorption of calcium before calcium phosphate, possibly less readily absorbed, may be formed; also they cite workers (49) who suggest that the specific organic phosphorus complex of casein reflects physiological needs peculiar to either the suckling young or to the processes of lactation. The McCollum group states:

> The nutritional significance of phosphoproteins has not received the critical attention this subject deserves. It does not seem probable that casein, for example, quantitatively the most important protein in the diet of all suckling animals, should contain approximately 0.8 per cent phosphorus in ester combination unless the particular complex is of nutritional importance and not merely a fortuitous circumstance. . . .

Casein, as it occurs in whole fluid milk, is accompanied by 2% or more of its own weight of calcium, with which it is chemically combined as calcium caseinate. About half of the total calcium of milk is said to be present as caseinate (50). The proportions of calcium contained in the casein of cheese are somewhat higher since the calcium (and also the phosphorus) is largely precipitated in the curd with the casein in the coagulation process (51). The occurrence with the phosphoprotein casein, in especially utilizable state, of calcium, the mineral which is regarded as the element in which diets are probably most often deficient, is a fortunate nutritional circumstance, of considerable importance in practical dietetics.

Lactalbumin Is Complete Protein of High Biological Value

Lactalbumin, although it occurs in milk in amounts (0.5%) considerably lower than does casein (3%), is an extremely important fraction of the milk protein system and has significant biological and nutritive properties.

TABLE 3

DAILY DIETARY ALLOWANCES FOR PROTEIN AS P

	DAILY PROTEIN ALLOWANCES RECOMMENDED BY COMMITTEE ON FOODS AND NUTRITION, NATIONAL RESEARCH COUNCIL		PERCENTAGES OF RECOMMENDED PROTEIN ALLOW PROVIDED BY UNIT AMOUNTS OF MILK AND MILK PRODUCTS					
	GRAMS OF PROTEIN	OUNCES† OF PROTEIN	FLUID MILK 1 Quart	EVAPORATED MILK 1 Ounce†	SWEETENED CONDENSED MILK 1 Ounce†	DRIED WHOLE MILK 1 Ounce†	DRIED SKIM MILK 1 Ounce†	CH AME CHE 1 Ou
Man (70 kg., or 154 lbs.):								
Moderately active	70	2.47	48.8%	2.8%	3.3%	10.4%	14.4%	9.
Very active	.....	.....	.....	.....	.....	.....	.....	.....
Sedentary	.....	.....	.....	.....	.....	.....	.....	.....
Woman (56 kg., or 123 lbs.):								
Moderately active	60	2.12	56.9%	3.3%	3.8%	12.2%	16.8%	11.
Very active	.....	.....	.....	.....	.....	.....	.....	.....
Sedentary	.....	.....	.....	.....	.....	.....	.....	.....
Pregnancy (latter half)	85	3.00	40.2%	2.3%	2.7%	8.6%	11.9%	8.
Lactation	100	3.53	34.1%	2.0%	2.3%	7.3%	10.1%	6.
Children up to 12 years:								
Under 1 year‡	3-4 g. per kg.	0.048-0.064 oz. per lb.	.....	.....	.....	.....	.....	.....
1-3 years§	40	1.41	85.3%	5.0%	5.8%	18.3%	25.2%	17.
4-6 years	50	1.76	68.3%	4.0%	4.6%	14.6%	20.2%	13.
7-9 years	60	2.12	56.9%	3.3%	3.8%	12.2%	16.8%	11.
10-12 years	70	2.47	48.8%	2.8%	3.3%	10.4%	14.4%	9.
Children over 12 years:								
Girls:								
13-15 years	80	2.82	42.7%	2.5%	2.9%	9.1%	12.6%	8.
16-20 years	75	2.65	45.5%	2.7%	3.1%	9.8%	13.5%	9.
Boys:								
13-15 years	85	3.00	40.2%	2.3%	2.7%	8.6%	11.9%	8.
16-20 years	100	3.53	34.1%	2.0%	2.3%	7.3%	10.1%	6.

‡Needs of infants increase from month to month. Amounts given are for approximately 6-18 months, and are expressed as grams per kilogram of body weight and as ounces per pound of body weight. Amounts of protein needed are less if from breast milk.

§Allowances are based on the middle age for each group (as 2, 5, 8, etc.) and for moderate activity.

ABLE 3.

N AS PROVIDED BY MILK AND MILK PRODUCTS*

ALLOWANCES MILK			APPROXIMATE AMOUNTS OF MILK AND MILK PRODUCTS PROVIDING TOTAL RECOMMENDED PROTEIN ALLOWANCES						
†	CHEESE AMERICAN CHEDDAR 1 Ounce†	ICE CREAM 1 Pint	FLUID MILK Quarts	EVAPORATED MILK Ounces†	SWEETENED CONDENSED MILK Ounces†	DRIED WHOLE MILK Ounces†	DRIED SKIM MILK Ounces†	CHEESE AMERICAN CHEDDAR Ounces†	ICE CREAM Pints
	9.7%	19.0%	2.1 qts.	35.3 oz.	30.5 oz.	9.6 oz.	7.0 oz.	10.3 oz.	5.2 pts.
	.....	.....	.....	.....	.....	.....	.....	.....	.....
	.....	.....	.....	.....	.....	.....	.....	.....	.....
	11.3%	22.1%	1.8 qts.	30.2 oz.	26.1 oz.	8.2 oz.	6.0 oz.	8.9 oz.	4.4 pts.
	.....	.....	.....	.....	.....	.....	.....	.....	.....
	.....	.....	.....	.....	.....	.....	.....	.....	.....
	8.0%	15.6%	2.5 qts.	42.8 oz.	37.0 oz.	11.6 oz.	8.4 oz.	12.6 oz.	6.3 pts.
	6.8%	13.3%	2.9 qts.	50.4 oz.	43.6 oz.	13.7 oz.	9.9 oz.	14.8 oz.	7.4 pts.
	.....	.....	0.04-0.05	0.69-0.92	0.59-0.79	0.19-0.25	0.14-0.18	.....	.....
	.....	.....	qts. per lb.	oz. per lb.	oz. per lb.	oz. per lb.	oz. per lb.	.....	.....
	17.0%	33.2%	1.2 qts.	20.2 oz.	17.4 oz.	5.5 oz.	4.0 oz.	6.0 oz.	3.0 pts.
	13.6%	26.5%	1.5 qts.	25.2 oz.	21.8 oz.	6.8 oz.	5.0 oz.	7.4 oz.	3.7 pts.
	11.3%	22.1%	1.8 qts.	30.2 oz.	26.1 oz.	8.2 oz.	6.0 oz.	9.0 oz.	4.4 pts.
	9.7%	19.0%	2.1 qts.	35.3 oz.	30.5 oz.	9.6 oz.	7.0 oz.	10.3 oz.	5.2 pts.
3									
	8.5%	16.6%	2.3 qts.	40.3 oz.	34.8 oz.	10.9 oz.	7.9 oz.	11.8 oz.	5.9 pts.
	9.0%	17.7%	2.2 qts.	37.8 oz.	32.7 oz.	10.3 oz.	7.4 oz.	11.1 oz.	5.5 pts.
	8.0%	15.6%	2.5 qts.	42.8 oz.	37.0 oz.	11.6 oz.	8.4 oz.	12.6 oz.	6.3 pts.
	6.8%	13.3%	2.9 qts.	50.4 oz.	43.6 oz.	13.7 oz.	9.9 oz.	14.8 oz.	7.4 pts.

*Milk proteins, in fluid milk, evaporated milk, sweetened condensed milk, dried whole milk, dried skim milk, cheese and ice cream, are all classified as proteins of the first class with equivalent values in the practical dietary.

†Ounces avoirdupois.

It is one of the most nutritively valuable proteins known, and its presence in milk supplements and enhances the already high biological value of casein.

Although early experiments resulted in some conflicting data regarding the biological value of lactalbumin in comparison with that of casein, it is now recognized that lactalbumin is fully as efficient as casein, and that under some conditions it has even higher biological value than casein.

Lactalbumin is similar in amino acid composition to blood serum albumin, although the two proteins are not identical and the elaboration of the albumin of milk is regarded as a distinct mammary synthesis rather than a mere transfer of albumin from the blood stream to the milk (52, 53, 54).

Following are some of the data obtained by various workers regarding lactalbumin and its amino acids:

The high leucine content of lactalbumin is similar to that of blood serum, but blood serum contains less glutamic acid than does lactalbumin (52).

Comparison of the amounts of sulphur-containing amino acids in the two chief proteins of milk show 0.37% cystine for casein and 3.1% for lactalbumin, and 3.2% methionine for casein and 2.8% for lactalbumin (9).

Growth experiments with rats fed casein and lactalbumin as the only source of protein in the diets showed that lactalbumin promoted better gains in weight per gram of protein intake than did casein (55, 56). Nitrogen storage was higher for lactalbumin than for casein (56). At 12% protein intake the biological value for lactalbumin was 80 and for casein 61. Paired feeding experiments showed that the same weight gain was obtained from one gram of lactalbumin nitrogen intake as from 1.33 grams of casein nitrogen intake. There was a greater nitrogen storage and less fat storage in rats fed lactalbumin as compared with those fed casein.

The casein requirement for normal growth in albino mice was found to be about 20% and the lactalbumin requirement about 12% (57).

The value of lactalbumin in human diets has been demonstrated. The addition of 50 to 100 g. daily of a cheese, containing lactalbumin instead of casein, to the diet of children two years of age and upward appeared to have a beneficial effect on the digestion and caused increased retention of nitrogen (58).

Lactalbumin Efficient in Correcting Low Blood Serum Albumin of Protein Deficiency

One of the deficiency diseases now attributed to severe protein deficiency is nutritional edema, accompanied by a marked lowering of the blood serum

protein. Decreased blood serum protein may result also from a pathological inability to generate plasma protein, as well as from dietary causes. Lactalbumin is a protein of superior value in promoting plasma protein regeneration in serum deficiencies resulting from either of these two causes[1]. Some of the data include:

> Lactalbumin is considered an efficient type of food for producing elevation of the serum albumin and is efficacious in the treatment of protein edema (59, 60). The accumulation of clinical evidence has shown that chronic depletion of serum albumin also is apt to be associated with disease of the liver (61).
>
> An assay method has been presented which appraises the potency of various food proteins with respect to their ability to promote the formation of serum albumin. Depletion of serum albumin is obtained in dogs by maintenance of a low protein diet; the regeneration induced by feeding a test food under standard conditions is measured. The potency value of a test food is the average of a number of assay values from experiments with different animals (62).
>
> Potency values for the relative efficiency of different proteins for synthesis of serum albumin, assigned after several series of tests, were reported (60) as follows:
>
> Beef serum (fresh) 0.801, beef serum (all assays) 0.770, lactalbumin 0.768, beef serum (dried) 0.739. Egg white 0.616, cow colostrum 0.611, egg (all assays) 0.608, egg (whole) 0.602. Milk 0.483, beef chuck 0.475, beef liver 0.445, casein 0.425.. Wheat gluten 0.207, gelatin -0.093.

Recommended Daily Protein Allowances Translated into Terms of Milk and Milk Products

The daily requirement for protein in the human diet ranges from about 1½ ounces to 3½ ounces. The daily allowances for persons of various ages, activity, and nutritive demand, as recommended to the National Nutrition Conference for Defense, by the Committee on Foods and Nutrition of the National Research Council (63), are shown in Table 3, translated into terms of milk and milk products.

The recommendations listed by the committee are for total protein, and do not include qualitative recommendations. Among the recommendations as to amounts of the proteins of animal origin that should be included in the diet, are those of the British Ministry of Health (64) and the British Medical Association's Committee on Nutrition (65) which are summarized by Boas Fixsen (66) in a review of the biological value of proteins:

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1. The subject of the dietary value of lactalbumin for elevation of blood serum albumin and for the correction of nutritional edema will be presented more fully in an early issue of *Borden's Review of Nutrition Research.*

> The unequal nutritive values of proteins are of great importance when it is sought to formulate standards for a complete human diet. To simplify their tasks, dietitians are accustomed to divide proteins into those of the first and second class, the first containing only those of animal origin, the second all others. How large should be the amount of first class protein in a diet has been the subject of controversy. The standard accepted by the Ministry of Health . . . was 37 g. of first class protein per day per man value, but the British Medical Association's Committee on Nutrition advocated the more generous figure of 50 g. . . .

Because of the high biological value of milk proteins, their superior supplementing qualities, their economy, and their unusual associations with minerals and vitamins, milk and milk products are among the preferred food sources from which much or all of the first class protein requirements can be met. The amounts of various milk products which will supply any proportion, or all, of 37 to 50 grams of protein per day can be calculated from the data in Table 3.

Milk Can Be Depended upon to Fill Entire Protein Need for Growing Children

The growing child has unusual protein requirements which can best be met by milk proteins. This is explained by Sherman (4):

> Growth at the normal rapid rate of early childhood involves the conversion of a very considerable part, sometimes as much as one third, of the protein of the food into body protein. This is accomplished to the best advantage when: (1) the protein of the food is largely of the kind most efficient in supporting growth, *i.e.*, milk protein; (2) the protein is well protected by the protein-sparing action of liberal amounts of carbohydrate and fat.
>
> . . . It is because the high protein requirement of childhood (for young children more than twice as much per unit of weight as for adults) is paralleled by an equally high energy requirement that the diet of the child need not contain a higher percentage of its calories in the form of protein than does the ordinary diet of the adult, if the protein for the child is well chosen.
>
> Usually, however, a well-planned dietary for a child will show a somewhat more than average proportion of its calories in the form of protein because after weaning the main feature of the child's diet should be cow's milk, which furnishes about 19 per cent of its calories in the form of protein. A child fed mainly upon cow's milk and taking enough food amply to cover his energy requirement will therefore receive a safe surplus of protein in highly efficient form. With an optimal amount of milk in the dietary of the growing child, the other foods may be selected with primary reference to other qualities than their protein content; without a liberal use of milk the proper feeding of a growing child becomes a very difficult problem, even more with respect to mineral and vitamin than to protein needs.

Milk Proteins Help Mothers Meet Extra Nutritional Demands

The human requirement for protein reaches its highest level during pregnancy and lactation. This is indicated in Table 3. Milk proteins, because of

their high biological value and because of their amino acid composition which is specialized for growth and which includes all the protein constituents which must be converted by the mother into milk for the infant, are of particular value in the maternal diet. The ability of milk proteins to facilitate milk production, more than that of other proteins, has been established by studies of lactation in both animal (67) and the human species (68). Physicians commonly recommend that milk and milk products should represent a major portion of the mother's diet.

Referring to the experiments cited on the value of milk proteins in promoting lactation, Sherman summarizes the data as follows:

> As in growth, so in lactation, the demand for material for the construction of new protein creates a condition in which differences of value in the protein fed may readily become more apparent than when only maintenance is involved. Hart and Humphrey found that in meeting the protein requirements of milch cows, milk protein and the protein of linseed meal are about 50 per cent more efficient than the proteins of the corn (maize) or of the wheat kernel; and Hoobler has shown that milk is an exceptionally good form of food protein for the production of human milk and the protection of the body protein of the nursing mother.

Milk as a Source of Superior Protein Obviously also Contributes Other Essential Dietary Factors

It is important to recognize that the chief energy foods, carbohydrates and fats, cannot fill the requirements for protein; fats may be metabolized into carbohydrates and carbohydrates into fats within certain limits but neither can be converted into protein. Protein, however, can contribute to the formation of body fat and carbohydrate, and it can also be utilized for muscular work and heat production. In addition, valuable amounts of calcium and phosphorus are chemically associated with the milk protein, casein. As is well known, milk and milk products are important sources of the vitamins. The non-fat portion of milk, in which the proteins occur, is a valuable dietary source of the water-soluble vitamins of the B complex group. Commercial casein has been shown to contain nutritionally significant amounts of B complex vitamins including riboflavin and the chick anti-dermatitis factor (69, 70); casein has experimentally demonstrable antirachitic properties (71) as have certain lactalbumins which are carriers of prosthetically bound pro-vitamin D (72).

In practice, the use of whole milk, skim milk, cheese, whey, ice cream and related products as protein sources also contributes incidentally, but in an important manner, to the intake of many other dietary requirements.

Milk Is One of Most Economical Protein Foods

Milk is important for its economy as a food source of proteins, as well as

for the superior nutritive quality of milk proteins. This important aspect of practical nutrition is summarized by Rose (73) who states:

> A quart of milk yields more than an ounce of pure protein of the highest quality, not only because of an assortment of essential amino acids whose efficiency in promoting growth is unexcelled, but also because under normal conditions it is the most completely digested and absorbed of all food proteins. From every point of view milk is an economical source of protein. It is produced at less expense than proteins of meat or eggs. An acre of cultivated farm land yields crops which when fed to farm animals give the following returns in human food:

Food Produced	Protein, Pounds	Total Calories
Milk	289	711,750
Beef	74	130,000
Mutton	59	137,295
Poultry and eggs	110	148,675

> Milk enhances the nutritive value of bread and other cereal proteins by adding those essential amino acids such as lysine and tryptophane in which cereal proteins are relatively poor. Experiments in feeding the lowest amount of protein capable of maintaining nitrogen balance in the adult have shown that less protein is required when milk is practically the sole source than when meat is so used and that proteins derived half from bread and half from milk furnish a mixture which is utilized with the same economy as milk alone.

Milk Products Reflect Economy of Milk as Protein Source in Daily Diet

This economy is reflected in the economy of milk products, an example of which is given by Wisconsin workers (74):

> . . . Because of its high protein content, cheese is an excellent source of this factor. It is inexpensive, too, compared with meat. One pound of American cheese at 35c a pound will supply 130 grams of protein of excellent quality, whereas one pound of beef-round at 40c supplies only 96 grams of protein. . . .

Similar Amino Acid Ratios of Human Milk and Cow's Milk Suggest Ideal Protein Pattern for Growth

Results of comparative analyses of samples of human and cow's milk have just been reported by Beach and associates (7). Similarities of the relative amino acid composition of the mixed proteins of the respective milks suggested to the research workers the possibility that the amino acid pattern of whole milk protein may be ideal for growth purposes and is therefore repeated in the two species.

The investigators reported the following wide differences in the two milks with respect to gross protein distribution:

> . . . Differences in the distribution are striking, human milk containing only 30 per cent as much total nitrogen and only 25 per cent as much protein nitrogen as does cow's milk. . . . In the two milks the whey protein occurs in approximately equal concentrations but the casein content of cow's milk is 8 times as great as that of human milk. 82 per cent of the protein nitrogen of cow's milk was found to be in the form of casein, while in human milk only 39 per cent was contained in the casein fraction, results which are similar to those found in the literature.

However, comparison of the amino acid compositions of the casein and whey protein fractions of the two milks indicated similarities:

> . . . The differences in amino acid composition are not great among the protein fractions of the two milks. The casein fraction of human milk has a significantly higher tryptophane content and is higher in cystine and lower in methionine than cow casein. The whey protein of human milks is significantly higher in arginine and cystine and lower in lysine and methionine than cow whey protein. . . .

To obtain a comparison of the relative amino acid compositions of the mixed proteins of cow and human milks, the research workers calculated their data on the basis of equal amounts of protein nitrogen. They tabulated the amino acid composition of 25.5 volumes of cow's milk, which contains a quantity of protein equal to that in 100 volumes of human milk. They discussed the comparative values as follows:

> . . . In spite of the differences between the two milks, in the distribution of protein nitrogen between casein and whey proteins, there is a surprising similarity in contour of the amino acid composition of the protein mixtures, with respect to histidine, arginine, lysine, tyrosine, and tryptophane. It is interesting to speculate whether the amino acid pattern of whole milk protein is ideal for growth purposes and therefore is repeated in the two species. The similarity in composition of the milk protein mixtures of the two species is not carried out with respect to cystine and methionine although the amounts of sulfur furnished by the proteins of the two milks are approximately equivalent.
>
> . . . In the proteins of cow's milk the preponderance of sulfur is in the form of methionine, with very little in the form of cystine, while in the proteins of human milk the sulfur is about equally divided between cystine and methionine.

Since maintenance of the adult may, in certain respects, be regarded as growth in a state of dynamic equilibrium in which a slower rate of tissue anabolism is approximately equalled by the daily rate of catabolism, the protein requirements of the adult might be expected to be qualitatively similar to those of the young; hence, the similarities of the protein pattern of the milk of the human species and of cow's milk, may in part explain the high nutritive value of milk proteins for individuals of all ages.

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